COMPARATIVE EXAMINATION OF A BACTERIUM PREPARATION (BACTOFIL® A10) AND AN ARTIFICIAL FERTILIZER [$CA(NO_3)_2$] ON CALCAREOUS CHERNOZEM SOIL

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SUMMARY

In a small-pot experiment a bacterium preparation, Bactofil® A10 and an artificial fertilizer containing $Ca(NO_3)_2$ in different dosages were studied on calcareous chernozem soil, concerning the readily available nutrient content of soil (nitrate-nitrogen, AL-phosphorus, AL-potassium content of soil, some soil microbial characteristics (total number of bacteria and fungi, cellulose-decomposing and nitrifying bacteria, CO_2-production of soil), and the amount of the plant biomass.

The readily available nutrient content of the calcareous chernozem soil increased due to the treatments, except for the change in the soil nitrate-nitrogen content, which did not measure up to the control due to the effect of high-dosage Bactofil.

The treatments also influenced the examined microbial characteristics of the soil positively. The artificial treatments significantly increased the total number of bacteria and the number of cellulose-decomposing and nitrifying bacteria. The low-dosage Bactofil significantly increased the number of cellulose-decomposing bacteria and both Bactofil dosage significantly increased the number of nitrifying bacteria. The measure of the soil respiration grew in all treatments, but significantly only in $Ca(NO_3)_2$ fertiliser treatments.

The quantity of the plant biomass was grew in a low-dosage Bactofil and significantly in the artificial fertiliser treatments. The highest plant biomass quantity was measured in the high-dosage artificial fertiliser treatment.

In the correlation analyses we found some medium positive correlation between the soil chemical, microbiological parameters examined, and the plant biomass in the case of both treatment-forms.

Based on our results $Ca(NO_3)_2$ artificial fertiliser treatments on calcareous chernozem soil proved to be more stimulating regarding the examined soil characteristics and the amount of the plant biomass, but the low-dosage Bactofil also positively influenced the majority of the soil characteristics examined in terms of nutrient supply.

Keywords: *organic fertilizer, Bactofil® A10, artificial fertilizer, $Ca(NO_3)_2$,, nutrient content, soil microbiology, plant biomass*

INTRODUCTION

Nowadays as opposed to modern cultivation and the agricultural activity, there is an increased expectation concerning environmental protection and „sustainable agriculture" which require the introduction of methods without the application of the chemicals- or with their reduction – that preserve or improve soil fertility (Kátai, 1997; Veres et al. 2007; Zsuposné, 2007).

Organic and green manures are among the oldest methods used for enriching the inorganic and organic colloid content of soils and for improving their water- and nutrient managements (Blaskó, 2005). Later, with intensive crop production, the application of artificial fertilizers started, which had deleterious effect (soil acidity) – primarily in the case of one-sided nitrogen artificial fertilisers – which unfavourably influence the biological activity of the soils, by this means the fertility of the soils and the expected quantity of yield (Lukácsné & Zsuposné, 2004).

Within integrated plant production there are many opportunities which may repair the fertility of soils with natural substances. According to Lazányi, (2003) there are three categories of natural soil amelioration materials: a., the group of the green manure which can be produced on the field and other organic matters b., those that get back into the soil as byproduct of the animal husbandry, such as livestock manure and compost, c., mined soil conditioning substances, like alginite (Solti, 1987), bentonite (Márton & Szabóné, 2002; Makádi et al., 2003; Szeder et al., 2008) or zeolite (Köhler, 2000).

By now the application of different bacterium preparations has become more frequent in agricultural production, particularly in the practice of „sustainable agricultural production" (Biró, 2006). Nowadays many researchers deal with the impact assessment of different microbiological preparations, both in field production and pot experiments under controlled conditions. Hereinafter we would mention some researchers:

Futó & Csorbai (2007) examined the effects of a soil bacterium product (Biorex) in maize and sunflower cultures on calcerous chernozem soil. They observed, that the soil bacterium product mobilize the natural nutrient supply of the soil and the average yield of sunflower increased by 20.4%, while that of maize by 17.14%. The condition and the resistance capacity of the plants improved by due to this preparation. They established that the preparation is suggestible for arable lands so as to achieve a well – balanced nutrient supply.

Makádi et al. (2007) studied the impacts of biogas-digestate and Phylazonit MC® on the green mass of second sowing silo maize (Zea mays L. „Coralba") and biological activity of soil. They applied the Phylazonit MC® treatment, the Phylazonit MC®+biogas-digestate treatments together, and the biogas-digestate treatment alone. The field experiment was set up at the region of Nyírbátor in 2007. The test plant was soybean (Glycine max L.). The Phylazonit MC®+biogas-digestate treatments together caused a significant increase in microbial activity compared to the control. On the basis of their statements, the applied treatments can be fitted with the sustainable agricultural practice successfully, especially on soils which have low humus content.

Gajdos et al. (2009) examined the effects of Phylazonit MC® on soil polluted with cadmium in the case of two plants and on the production and nutrient uptake of the sunflower (Helianthus annus L. cv. Arena PR) and maize (Zea mays L. cv. Norma SC). They observed that the cadmium accumulated in the root primarily. The sunflower absorbed more cadmium, and it had presumably larger stress-tolerance than maize. They experimented with the use of a bacterium-containing biofertilizer and the toxic effect of cadmium was moderated.

Kincses et al. (2007) studied the effects of bacterium manure on the biomass of the perennial rye-grass on two soil types. Their results – planning the achievement of an identical crop level too – with the allocation of stem residues and bacterium manure into the soil can be reducible the using of the quantity of artificial fertiliser, reducing the environmental loading. The effects of the bacterium manure extended over the following season too and its effect intensified.

Balláné et al. (2007) examined the effects of an artificial fertilizer and a bacterium manure in lettuce culture (Lactuca sativa L.) on calcareous chernozem and humus sandy soil. The nitrogen artificial fertiliser significantly increased the wet and dry biomasses of the plant, and advantageously influenced the nitrate and all-nitrogen contents of the plant. In the case of the application of the bio-manure (Phylazonit MC®) the effect was greatly influenced by soil characteristics. A no significant change was experienced in the production of plant dry matter. On the chernozem soil the all-nitrogen and nitrate-contents increased, while a small-scale change was experienced in the measured parameters on sandy soil.

Kincses et al. (2008) examined the effects of artificial, organic and bacterium manures on the 0.01 M $CaCl_2$-solube N-, P- and K contents of soils, and on the change in the acidity of the soils. Based on their results, they established that the organic fertiliser was able to increase the pH of the acidic sandy soil. The combined N-dressing (artificial and organic manure) provided N -, P- and K to the plants under the whole season, while the soil did not became acid. The favourable effects of bacterium manuring for plants readily available N-, P-, K-contents was observed only on chernozem soil.

The aim of our examination was to study the effect of a bacterium preparation, Bactofil® A10, and an artificial fertilizer, $Ca(NO_3)_2$ in different dosages on calcareous chernozem soil on the readily available (AL-solube) nutrient content of soil, some soil microbial characteristics, and on the amount of the plant biomass.

MATERIALS AND METHODS

The pot experiment was set up at the greenhouse of the UD CAAES, Department of Agrochemistry and Soil Science on calcareous chernozem soil ($pH_{(H2O)}$ 7.47) in 2007-2009. The experiment was performed in three repetitions with one kg soil per pot. The moisture content of soils was set at 70% of maximum water holding capacity. As a test plant, perennial ryegrass *(Lolium perenne L.)* was used. Samples were collected in the fourth and eighth weeks after the emergence of the test plant.The grass was cut by scissors. The biomass of the test plant was also measured every sampling time. The plant biomass after the cutting got into the drying cupboard, then the the dry matter was measured. The plant production was the sum of cuttings of the two sampling dates in every year. After the second sampling the experiment was terminated. After soil homogenization, the laboratory examinations were performed at the soil chemistry and soil microbiology lab of the Department. As a basic treatment, 100 mg P_2O_5 and 100 mg K_2O were applied in each pot as a common solution of potassiumdihydrogen-phosphate and potassium-sulphate were applied. The nitrogen was added into the soil in form $Ca(NO_3)_2$ solution.

We made three treatments in 2007, while in 2008-2009 complementing growing dosages were given out. The different treatments are presented in *Table 1.*

Table 1

The Bactofil and $Ca(NO_3)_2$ treatments and dosages applied

Treatments number	Ttreatments and dosages	
	2007.	2008-2009.
1.	Control	Control
2.	* Bactofil® A10	* Bactofil® A10
3.	-	** Bactofil® A10
4.	100 mg kg^{-1} N [$Ca(NO_3)_2$]	100 mg kg^{-1} N [$Ca(NO_3)_2$]
5.	-	200 mg kg^{-1} N [$Ca(NO_3)_2$]
** 2.5 times the field dosage (10,75*10^5bacteria kg^{-1})*		
*** 5 times the field dosage (21,50*10^5bacteria kg^{-1})*		

We measured the nitrate-N content with the methode of Na-salicilate (Felföldy, 1987), the readily available phosphorus and potassium contents by means of extraction of ammonium lactate-acetate. The AL-P_2O_5 with spectrophotometer, the AL-K_2O with flamephotometer were measured (Gerei, 1970). From among the microbial parameters, the total number of bacteria and microscopic fungi were determined by plate dilution from soil-water suspension (on Bouillon soup, and peptone-glucose agar) (Szegi, 1979). The number of cellulose-decomposing and nitrifying bacteria was determined by most probable number of bacteria method according to Pochon & Tardieux, (1962). We also measured the amount of carbon-dioxide released from the soil in 10 days (Witkamp, 1966 cit. Szegi, 1979) and the biomass of the test plant was measured.

The results were evaluated statistically, the means of samplings, deviation and significant differences *(p=5)* were calculated and correlation analysis was applied for revealing the relationships between the studied parameters. The statistical evaluation was done using the SPSS 13.0 programme.

RESULTS AND DISCUSSION

The effects of the treatments on the examined soil characteristics are presented separate in each year (2007-2009), the tables and figures include also the averages of the repetitions at the two sampling dates in 2007-2009. In the publication the means of the results of the three examination years come to analyse.

The readily available nutrient content of soil *(Table 2)* grew due to the treatments in the case of all examined parameters, except for the change in the soil nitrate-nitrogen content, which did not measure up to the control on the effect of high-dosage Bactofil. Both dosages of artificial fertilizer significantly increased the readily available nutrient content of soil. The higher-dose of the artificial fertiliser proved to be more stimulating in the case of all examined parameters.

Table 2

The effect of the different nutrient supplying forms on the readily available nutrient content of soil (2007-2009.)

Treatments number	Nitrate-N (mg $1000g^{-1}$)				AL-P_2O_5 (mg $1000g^{-1}$)				AL-K_2O (mg $1000g^{-1}$)			
	2007.	2008.	2009.	Means	2007.	2008.	2009.	Means	2007.	2008.	2009.	Means
1	5.29	5.98	7.01	6.09	182.67	192.67	178.33	184.56	373.33	328.67	318.33	340.11
2	*4.26	*6.68	7.53	6.16	192.00	*214.67	187.67	198.11	*353.33	*340.00	*362.08	351.80
3	-	6.30	4.68	5.49	-	203.17	188.28	195.72	-	*315.00	*378.75	346.88
4	*6.18	*6.95	*8.21	*7.11	*209.67	*217.83	188.00	*205.17	383.33	*348.00	*376.25	*369.19
5	-	*7.62	7.39	*7.51	-	*211.50	*207.89	*209.69	-	*364.17	*384.58	*374.38
*$LSD_{5\%}$	0.46	0.61	0.76	0.94	14.92	16.50	12.95	14.46	14.17	10.00	13.16	12.78

Among the soil microbial parameters *(Table 3)*, the total number of bacteria was measured in both treatment types. Their number was significantly raised by artificial fertilizer treatments, but we were not able to demonstrate a significant difference between the dosages.

The number of microscopic fungi did not change significantly compared to the value of the control, and there were no significant differences between the treatments.

Table 3

The effect of the different nutrient supplying forms on the total number of bacteria and fungi (2007-2009.)

Treatments number	Total number of bacteria ($^{X}10^6g^{-1}$ soil)				Total number of fungi ($^{X}10^3g^{-1}$ soil)			
1	5.30	4.03	8.07	5.80	40.33	53.00	79.83	57.72
2	*6.02	5.19	7.33	6.18	45.50	56.33	73.50	58.44
3	-	4.98	7.25	6.12	-	51.83	*65.33	58.58
4	*7.95	*5.42	*5.84	*6.40	*65.67	*58.67	*47.00	57.11
5	-	*6.12	7.40	*6.76	-	*61.83	*56.50	59.17
*$LSD_{5\%}$	0.60	1.24	1.73	0.52	6.03	5.33	12.72	5.91

The amount of the two physiological groups, the number of cellulose-decomposing and nitrifying bacteria *(Table 4)* also increased compared to the control. The amount of cellulose-decomposing bacteria changed more than two and a half-fold, while the number of nitrifying bacteria changed nearly fourfold. The low-dosage Bactofil and the artificial fertilizer treatments significantly raised the number of the cellulose-decomposing bacteria, while in the case of the nitrifying bacteria both the Bactofil and the artificial fertiliser treatments proved to be stimulating. A significant difference was observed between the two treatments in the case of the two physiological groups, the high-dosages artificial fertiliser treatments were more stimulating.

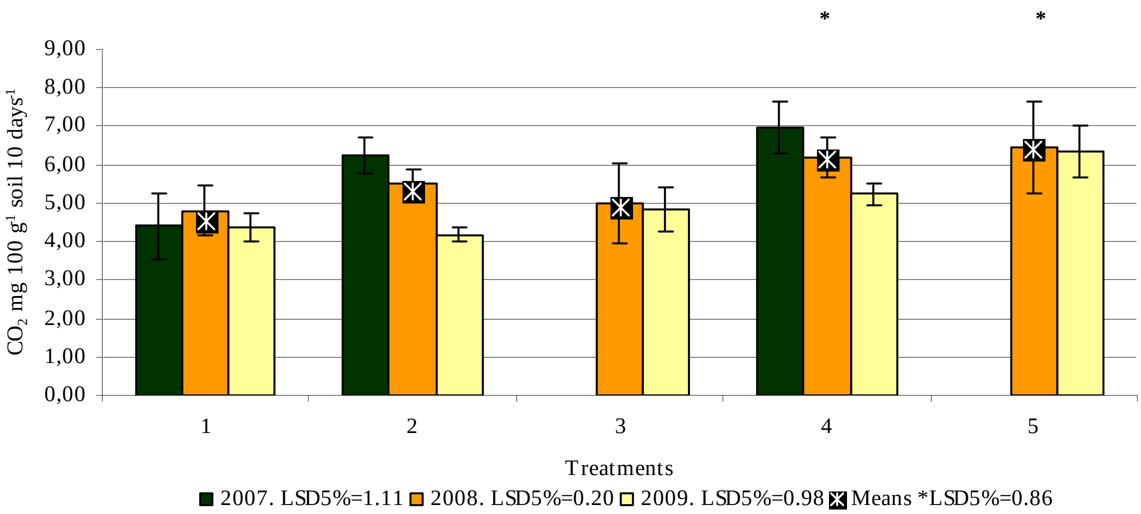
Table 4

The effect of the different nutrient supplying forms on the amount of cellulose-decomposing and nitrifying bacteria (2007-2009.)

Treatments number	Cellulose-decomposing bacteria ($^{X}10^3 g^{-1}$ soil)				Nitrifying bacteria ($^{X}10^3 g^{-1}$ soil)			
1	8.50	5.55	2.20	5.42	3.35	3.75	2.19	3.10
2	*14.08	*8.40	*21.45	*14.64	*10.35	6.67	3.80	*6.94
3	-	*8.35	*9.92	9.14	-	7.37	*6.91	*7.14
4	11.58	6.65	*19.49	*12.57	4.35	*11.08	*6.20	*7.21
5	-	6.40	*23.00	*14.70	-	*15.33	*7.65	*11.49
*$LSD_{5\%}$	5.18	1.19	7.41	4.26	5.31	5.07	2.63	2.34

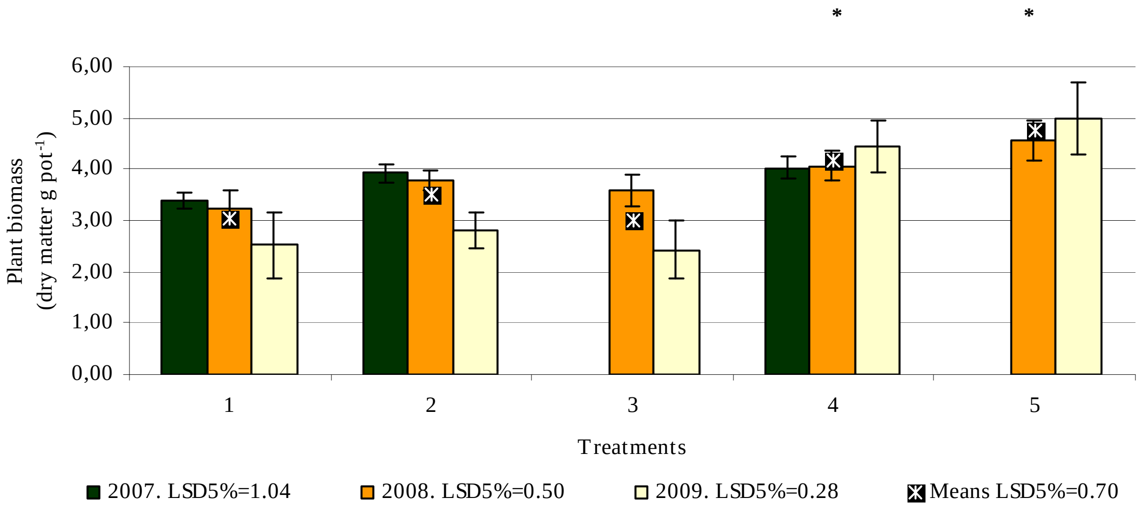
The measure of the soil respiration *(Figure 1)* grew in all treatments, but the growth was significant only in $Ca(NO_3)_2$ fertiliser treatments.

Figure 1: **The effect of the different nutrient supplying forms on CO_2-production of soil (2007-2009.)**



The quantity of the plant biomass *(Figure 2)* grew both in the small-dosage Bactofil and in the artificial fertiliser treatments. The growth was significant in the artificial fertiliser treatments, but the difference was not significant between the dosages. The quantity of the plant biomass was nearly to the control due to the high-dosage Bactofil.

Figure 2: **The effect of the different nutrient supplying forms on the amount of the plant biomass (2007-2009.)**



The results were evaluated by correlation analysis to determine the relationship between the changes in the soil nutrient content, the microbial parameters studied, and plant biomass in the Bactofil and $Ca(NO_3)_2$ artificial fertilizer treatments respectively *(Table 5)*.

We found some medium positive relationships between the studied soil parameters and the plant biomass in both treatment forms. We could not determine a strong correlation in the case of either treatments.

In the Bactofil treatments medium positive correlations were found between the nitrate-N content of soil and the amount of microscopic fungi (r=0.553), the AL-solube phosphorus and the amount of plant biomass (r=0.542), the AL-potassium and the number of nitrifying bacteria (r=0.539), the total number of bacteria and the CO_2-production of soil (r=0.615), the amount of microscopic fungi and soil respiration (r=0.685), and between the number of cellulose-decomposing bacteria and plant biomass (r=0.717).

In the $Ca(NO_3)_2$ artificial fertilizer treatments medium positive correlations were found between the nitrate-N content and the amount of plant biomass (r=0.527), the AL-solube phosphorus and cellulose-decomposing bacteria (r=0.650) and plant biomass (r=0.513). Further medium correlations were between the AL-potassium content of soil and the number of nitrifying bacteria (r=0.606) and the amount of plant biomass (r=0.599), between the cellulose-decomposing bacteria and the plant biomass (r=0.580), and between the amount of plant biomass and soil respiration (r=0.620).

Table 5

Relationships between the soil parameters examined and the plant biomass (2007-2009.)

[x]Soil parameters	Pearson correlation								
	Bactofil treatments (by treatments 1; 2 n=18) (by treatment 3 n=12)								
	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.
I.	1								
II.	0.122	1							
III.	0.102	0.046	1						
IV.	0.329	0.135	0.110	1					
V.	0.553(**)	0.052	0.169	0.452	1				
VI.	0.110	0.404	0.110	0.075	0.040	1			
VII.	0.154	0.120	0.539(**)	0.144	0.218	0.344	1		
VIII.	0.287	0.019	0.316	0.615(**)	0.685(**)	0.272	0.417	1	
IX.	0.148	0.542(**)	0.079	0.065	-0.173	0.717(**)	0.123	0.000	1
	$Ca(NO_3)_2$ artificial fertilizer treatments (by treatments 1; 4 n=18) (by treatment 5 n=12)								
	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.
I.	1								
II.	0.189	1							
III.	0.229	0.340	1						
IV.	0.409	0.187	0.212	1					
V.	0.351	0.189	-0.029	0.498	1				
VI.	0.368	0.650(**)	0.218	0.010	0.101	1			
VII.	0.385	0.146	0.606(**)	0.189	-0.201	0.116	1		
VIII.	0.497	0.281	0.447	0.480	0.497	0.266	0.451	1	
IX.	00.527(**)	0.513(**)	0.599(**)	0.442	0.189	0.580(**)	0.483	0.620(**)	1
** Correlation is significant at the 0.01 level (2-tailed).									

[x] I.Nitrate-N; II.AL-P_2O_5; III.AL-K_2O; IV.Total number of bacteria; V.Total number of fungi; VI.Cellulose-decomposing bacteria; VII.Nitrifying bacteria; VIII. CO_2-production of soil; XI.Plant biomass

CONCLUSIONS

- The readily available nutrient content of the calcareous chernozem soil increased due to the treatments, except for the change in the soil nitrate-nitrogen content, which did not measure up to the control on the effect of high-dosage Bactofil. In point of the readily available nutrient content of soil, the artificial fertiliser treatments proved to be more stimulating.
- The treatments positively influenced the examined microbial characteristics of the soil. The artificial treatments raised the total number of bacteria, the number of cellulose-decomposing and nitrifying bacteria significantly. The low-dosage Bactofil significantly increased the number of cellulose-decomposing bacteria and both Bactofil dosages significantly raised the number of nitrifying bacteria. The measure of the soil respiration grew in all treatments, but the growth was significant only in $Ca(NO_3)_2$ fertiliser treatments.
- The quantity of the plant biomass grew in the low-dosage Bactofil and significantly in the artificial fertiliser treatments. The highest plant biomass quantity was measured in the high-dosage artificial fertiliser treatment.

- In the correlation analysis we have found some medium positive correlations between the soil chemical, microbiological parameters examined, and the plant biomass in the case of both treatment-forms.
- Based on our research $Ca(NO_3)_2$ artificial fertiliser treatments on calcareous chernozem soil proved to be more stimulating regarding the examined soil characteristics and the quantity of the plant biomass. The artificial fertlizer treatments was more efficient in case of the nutrient supply of soil with six percent (6.15%), the examined soil microbiological characteristics with forty percent (in case of the total number of bacteria with 10.52%, the number of cellulose-decomposing bacteria with 32.10%, and nitrifying bacteria with 74.52%). The CO_2-production of soil was with thirty (25.66%), the amount of plant biomass was with fourty (39.87%) percent higher in $Ca(NO_3)_2$ treatments then in Bactofil treatments. However the low-dosage Bactofil influenced positively the majority of the soil characteristics examined, particularly in case of the nutrient supply.

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